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# Continuous lighting improves the leaf quality of sweet basil (*Ocimum basilicum* L.) grown in a controlled environment

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All relevant data are within the paper and its Supporting Information files.

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**Abstract: In vertical farms, continuous lighting (CL) with lower light intensity (photosynthetic photon flux density, PPFD) is a method to reduce the investment costs for the lighting system. Continuous lighting has both negative and positive effects on crop performance, depending on the plant species. In this study, we investigated the effect of CL on plant growth and leaf quality in sweet basil (*Ocimum basilicum* L. cv. Tigullio) cultivated in a growth chamber with light emitting diode (LED) (R:B:G=3:1:1). Basil plants were grown hydroponically for 14 days with a photoperiod of 16 h $d^{-1}$ (control) or 24 h $d^{-1}$ with a PPFD of 220 or 147 µmol $m^{-2}$ $s^{-1}$, respectively. The daily light integral was 12.7 mol $m^{-2}$ $d^{-1}$ in both treatments. Plant growth was not significantly affected by the light regime. Compared with the control, CL increased the leaf antioxidant capacity and concentration of total chlorophylls, flavonoids and phenols, and reduced the nitrate level. Continuous lighting would slightly increase or decrease electricity costs compared to 16-hour illumination, depending on the daily schedule of the standard lighting regime.**

## 1. Introduction

In many countries, high-value fresh vegetables and herbs are increasingly produced in plant factories with artificial light, also known as vertical farms. In vertical farms, crop yield is enhanced due to the use of multi-tiered cultivation systems and the maintenance of optimal environmental conditions for plants (Zhu and Marcelis, 2023). In a controlled environment, plant growth is primarily determined by the daily light integral (DLI) (Warner *et al.*, 2023), which is the light level plants receive over a day and thus depends on the photosynthetic photon flux density (PPFD) and the photoperiod. Generally, crop yield and quality are boosted by increasing DLI to a certain level. In vertical farms, typical DLI values are 12 to 17 mol $m^{-2}$ $d^{-1}$ for leafy greens and herbs, and 15 to 40 mol $m^{-2}$ $d^{-1}$ for fruiting crops (MechaTronix Horticulture Lighting, 2025). The photoperiod is generally 16 or 18 h $d^{-1}$ with PFFD ranging between 150 and 300 µmol $m^{-2}$ $s^{-1}$) (Nájera *et al.*, 2022).

The main drawback of vertical farms is the high electricity requirement for artificial illumination and air conditioning (Zhu and Marcelis, 2023). Despite the adoption of energy-efficient light-emitting diode (LED) fixtures, the lighting system remains one of the main components of the capital costs of vertical farms and accounts for approximately 80% of the total electricity demand (Cai *et al.*, 2024). Therefore, the key to reducing energy costs and/or enhancing energy use efficiency in vertical farms is innovating lighting systems and strategies (Liu *et al.*, 2022).

Continuous lighting (CL; i.e., 24-hour photoperiod without dark interruption) with lower PFFD is a cost-effective strategy to achieve the same DLI in vertical farms, as it reduces the investment costs for the lighting system (fewer fixtures are necessary) and the operational costs for illumination in case of a time-based pricing scheme for electricity (Lanoue *et al.*, 2022).

Long photoperiod (>18 h $d^{-1}$) can enhance plant production by increasing leaf photosynthesis. Under the same DLI, longer photoperiods with lower PPFD increase yield in most species (Warner *et al.*, 2023), since crop light use efficiency rises with decreasing irradiance (e.g., Palmer and van Iersel, 2020; Carotti *et al.*, 2021). This explains why many studies have been conducted on the effect of CL in greenhouse crops, which started nearly one century ago (Arthur *et al.*, 1930), and more recently in vertical farms (Velez-Ramirez *et al.*, 2011; Shibaeva *et al.*, 2023 a). However, leaf injury and growth inhibition were observed in some species grown under greenhouse conditions with CL for a relatively long period, such as tomato, eggplant, pepper, and cucumber (Velez-Ramirez *et al.*, 2011; Shibaeva *et al.*, 2023 a). Although the mechanism of leaf damage under CL has not yet been completely elucidated, it has been attributed to carbohydrate accumulation in leaf tissues and disruption of endogenous circadian rhythms, which results in early senescence and reduced photosynthesis (Velez-Ramirez *et al.*, 2011; Shibaeva *et al.*, 2023 a). In contrast to fruiting crops, leafy vegetables and herbs have a short growing cycle and generally, they are not negatively affected by long photoperiod or CL (Shibaeva *et al.*, 2023 a); positive effects have also been reported (Table 1S). For instance, in rocket CL enhanced plant growth and improved leaf quality by reducing nitrate concentration and increasing the concentration of antioxidants (Proietti *et al.*, 2021).

Sweet basil (*Ocimum basilicum* L.) is an economically important herb cultivated worldwide (Camlica and Yaldiz, 2023) due to its adaptability to different growing conditions and systems. Its short cycle, rapid growth, and easy cultivation make basil one of the most used species in vertical farms; basil is the most studied species for controlled environment agriculture after lettuce (Dsouza *et al.*, 2023). There exists a reasonable consensus on the optimal DLI for basil grown indoors, which ranges between approximately 13 and 15 mol $m^{-2}$ $d^{-1}$ (Liaros *et al.*, 2016; Dou *et al.*, 2018; Pennisi *et al.*, 2020), but contrasting results have been found on the response of this species to CL, since positive (Islam *et al.*, 2010; Lanoue *et al.*, 2022; Fayezizadeh *et al.*, 2024), negative (Beaman *et al.*, 2009), or no (Pennisi *et al.*, 2020) effects have been reported.

In this work, the impact of CL on plant growth and leaf quality was studied in sweet basil cultivated hydroponically in a growth chamber with LED light under a photoperiod of 16 or 24 h $d^{-1}$ with the same DLI (12.7 mol $m^{-2}$ $d^{-1}$). Based on the results in the literature and a preliminary experiment, in which only growth parameters were measured, we hypothesized that CL does not affect plant growth but improves leaf quality by reducing nitrate accumulation and increasing the concentration of antioxidant compounds.

## 2. Materials and Methods

*Plant material and growing conditions*

Two experiments were conducted with sweet basil (cv. Tigullio; Franchi Sementi, Grassobbio, Italy) grown in a floating raft system in a growth chamber at the University of Pisa.

Basil seeds were sown in 240-cell trays with stone wool plugs and 41 days after sowing, the seedlings with two pairs of true leaves were transplanted in plastic tanks with 14 litres of aerated nutrient solution. The solution had a pH of 6.0 and an electrical conductivity of 2.39 dS $m^{-1}$, and contained the following concentration of nutritive elements: N-$NO_3$ 10.0 mM, P 1.5 mM, K 9.0 mM, Ca 4.5 mM, Mg

2.0 mM, Fe 40.0 µM, B 40.0 µM, Cu 3.0 µM, Zn 10.0 µM, Mn 10.0 µM, and Mo 1.0 µM. Twelve plants were grown in each tank and crop density was approximately 96 plants $m^{-2}$. Air temperature and relative humidity were kept at 24.0°C and 65% - 70%, respectively.

Basil plants were illuminated by red, blue, and green (R:B:G=3:1:1) (Fig. 1) LED tubes (Circular Natural Indoor, C-Led, Imola, Italy) with a 16-hour photoperiod at 220 µmol $m^{-2}$ $s^{-1}$ PPFD, till transplantation in hydroponic tanks. Afterwards, one group of plants was kept under the same light regime (control) while another group was grown with a 24-hour photoperiod at 147 µmol $m^{-2}$ $s^{-1}$ PPFD (CL). The lowest PPFD was achieved by using fewer lamps and slightly adjusting the distance between lamps and the plant canopies. An opaque plastic screen was used to avoid contamination between the two light treatments, which lasted 14 days. Each treatment had four replicates, each consisting of one hydroponic tank.

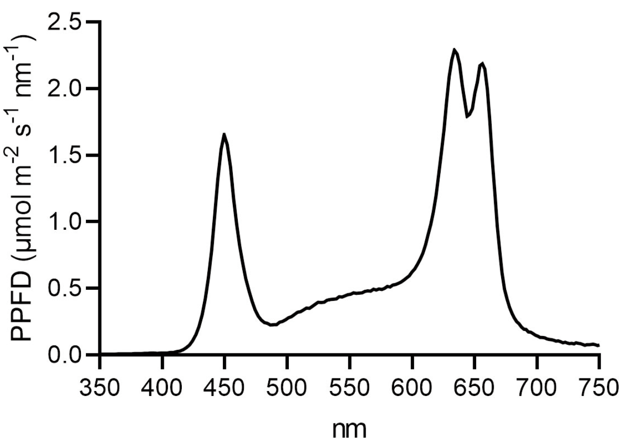


Fig. 1 - The spectral distribution of the LED tubes (Circular Natural Indoor, C-Led, Imola, Italy) used in the experiments with sweet basil in a growth chamber. The spectrum was measured using a portable spectroradiometer (SpectraPen LM 510, Photon Systems Instruments, Drásov, Czech Republic).
PPFD= photosynthetic photon flux density.

*Determinations*

Leaf fresh (FW) and dry weight (DW), stem and root DW, plant height, and leaf area were measured in plant samples collected in each replicate; each sample consisted of six individual plants. Dry weight was determined in plant samples that were dried in a ventilated oven at 70°C until they were of constant weight. Leaf area was measured using a digital planimeter (DT Area Meter MK2, Delta T-Devices). Leaf area index (LAI) was computed as the average leaf area per plant divided by the area occupied by one plant.

Twelve days after the beginning of the experiment, stomatal behaviour was assessed by measuring the abaxial leaf diffusion conductance ($g_s$) with a diffusion porometer (AP4, Delta-T Devices, Cambridge, UK). The $g_s$ was measured on two individual plants in each replicate of both treatments at two times corresponding to midday and midnight of the control treatment.

Plant water uptake was determined by measuring the weight of each hydroponic tank at the beginning and the end of each experiment. The weight difference was assumed to be equal to the plant water absorption because the tank was covered by the polystyrene floating raft and thus direct evaporation was insignificant.

Leaf concentration of mineral elements was determined in dried samples whereas the antioxidant capacity and the concentration of total chlorophylls, carotenoids, flavonoids, and phenols were examined in fresh samples. Each sample comprised all leaves from six individual plants collected from each tank.

To determine leaf mineral concentration, finely ground samples underwent mineralization in a 5:2 v/v mixture of 65% $HNO_3$ and 35% $HClO_4$ at 240°C for one hour or were extracted with distilled water at room temperature for two hours. The mineralized samples were utilized for assessing the concentrations of K, Ca, Mg, Cu, Fe, Mn, and Zn through atomic absorption spectroscopy. At the same time, P levels were determined spectrophotometrically with Olsen's method. Water extracts were analysed spectrophotometrically for nitrate concentration using the salicylic-sulfuric acid method (Puccinelli *et al.*, 2023).

Methanol (99% v/v) was used to extract fresh samples, followed by a 60-minute sonication (frequency 28-34 kHz, power peak 350 W). The samples were stored at -18°C for 24 hours; afterwards, the concentrations of total chlorophylls, carotenoids, phenols, and flavonoids were determined spectrophotometrically as previously reported (Puccinelli *et al.*, 2023). The antioxidant capacity of leaf samples was measured in methanol extracts using the ferric-reducing ability of plasma (FRAP) assay (Benzie and Strain, 1996) and the 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay (Blois, 1958).

*Electricity costs of light regimes*

The electricity cost of the light regimes tested in this work was calculated every week using the

scheme for time-of-use pricing of electricity in place in Italy, which differentiates the prices by single-hour rate and time slot (F), as follows (ENEL, 2025 a):
F1: from 8.00 am to 7.00 pm from Monday to Friday, excluding national holidays;
F2: from 7.00 am to 8.00 am and from 7.00 pm to 11.00 pm from Monday to Friday, Saturday from 07.00 am to 11.00 pm, excluding national holidays;
F3: from 11.00 pm to 7.00 am from Monday to Saturday, Sunday, and national holidays.

The prices used for calculation were 0.1783, 0.1716 and 0.1485 € kWh$^{-1}$ for F1, F2, and F3, respectively. They are mixed costs because they have variable and fixed components. The variable component is the PUN Index GME (PUN is the Italian acronym for Prezzo Unico Nazionale, "National Single Price"), which is the weighted average of electricity prices in different areas of Italy and at different times of the day. The fixed part is the contribution paid to the energy provider. We used a fixed component of 0.020 € kWh$^{-1}$ (ENEL, 2025 a) while the PUNs were the mean values recorded in January 2025 for the three time slots (ENEL, 2025 b).

The weekly lighting cost was computed for three light regimes with the same DLI (12.7 mol m$^{-2}$ d$^{-1}$): 220 µmol m$^{-2}$ s$^{-1}$ PPFD for 16 h d$^{-1}$ from 11:00 pm until 03:00 pm of the next day (LR1) or from 08:00 am until 12:00 pm (LR2); 147 µmol m$^{-2}$ s$^{-1}$ PPFD for 24 h d$^{-1}$ (LR3). The consumption of electricity was estimated using a photon efficacy of LED lamps of 3.1 µmol J$^{-1}$.

*Statistical analysis*

Since the two experiments were conducted in the same growing conditions and the results were quite similar, the data were pooled, subjected to 1- or 2-way ANOVA, and reported as the mean values (±SE) of eight replicates. Leaf diffusion conductance data were subjected to 2-way ANOVA, with the time of measurement and light regime as variability factors. Data were tested for the normality of the distribution using the Shapiro-Wilk test and for the homogeneity of variances using Levene's test. Statistical analysis was performed using JMP statistical software.

## 3. Results

*Plant growth*

In our study, the light regime did not affect basil growth as no significant differences (P>0.050) were found between the controls and CL plants regarding plant height, leaf and stem FW (yield), and leaf, stem, root, and total DW (Table 1). The production for the market of bunched fresh herbs was 0.96 kg m$^{-2}$, a noticeable yield for a crop that lasted only two weeks. The leaf area index has an average value of 2.16 and was not significantly affected by the light regime (data not shown)

*Plant water and mineral relations*

The cumulated water uptake during the experiments was not significantly influenced by the light regime and averaged 34.8±2.1 L m$^{-2}$. This result is consistent with the absence of significant differences in LAI between the controls and CL plants, and the measurements of $g_s$, which did not vary significantly between the two plant groups at both midday and midnight of the control light regime

Table 1 - Leaf and stem fresh (FW) and dry weight (DW), root and total DW and plant height, in sweet basil plants grown hydroponically for 14 days in a growth chamber under two LED light regimes with the same daily light integral (12.7 mol m$^{-2}$ d$^{-1}$): a 16-hour photoperiod and 220 µmol m$^{-2}$ s$^{-1}$ PPDF (Control); 24-hour photoperiod and 147 µmol m$^{-2}$ s$^{-1}$ PPDF (CL)

| | Light regime | | ANOVA significance |
|---|---|---|---|
| | Control | CL | |
| Leaf FW (kg m$^{-2}$) | 0.783±0.044 | 0.748±0.005 | NS |
| Stem FW (kg m$^{-2}$) | 0.198±0.018 | 0.195±0.012 | NS |
| Leaf DW (kg m$^{-2}$) | 0.059±0.003 | 0.058±0.003 | NS |
| Stem DW (kg m$^{-2}$) | 0.015±0.004 | 0.015±0.004 | NS |
| Root DW (kg m$^{-2}$) | 0.013±0.001 | 0.012±0.001 | NS |
| Total DW (kg m$^{-2}$) | 0.087±0.004 | 0.084±0.004 | NS |
| Plant height (cm) | 11.9±0.2 | 11.5±0.3 | NS |

Mean values (±SE) of eight replicates.
PPFD= photosynthetic photon flux density. Significance level: NS = not significant.

(Fig. 2). However, on average $g_s$ was significantly higher at midday (290.0±8.6 mmol $H_2O$ $m^{-2}$ $s^{-1}$) that at midnight (187.7±12.1 mmol $H_2O$ $m^{-2}$ $s^{-1}$) .

In both experiments, no plant revealed evident mineral deficiency symptoms and the leaf concentration of nutritive elements (Table 2) was within or above the adequate levels reported for sweet basil (Bryson *et al.*, 2014). Nonetheless, CL plants showed a significantly lower leaf concentration of Mg and Mn, and a higher concentration of Zn than the control plants. In Japanese mugwort, CL induced a lower leaf concentration of K, Mn, and Zn, in partial agreement with our findings (Hata and Kawamura, 2023).

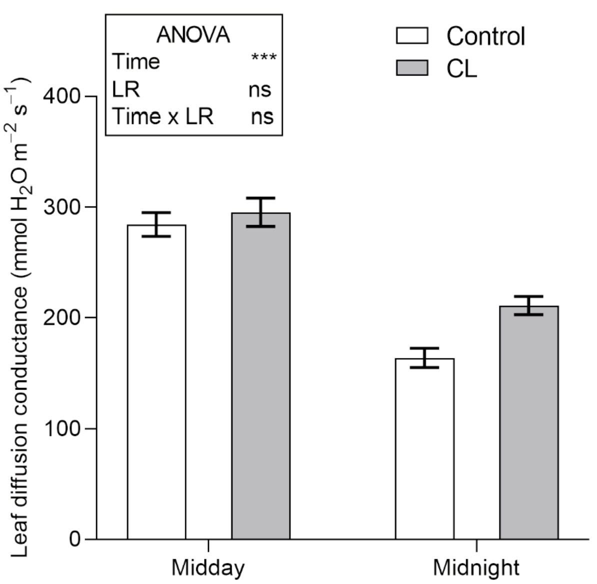


Fig. 2 - Abaxial leaf diffusion conductance in sweet basil plants grown hydroponically for 14 days in a growth chamber under two LED light regimes (LR) with the same daily light integral (12.67 mol $m^{-2}$ $d^{-1}$): a 16-hour photoperiod and 220 µmol $m^{-2}$ $s^{-1}$ PPDF (Control); 24-hour photoperiod and 147 µmol $m^{-2}$ $s^{-1}$ PPDF (CL). The measurements were taken at midday and midnight of the control light regime 12 days after the beginning of the experiment. Mean values (±SE) of eight replicates. Significance level: *** P ≤ 0.001; ** P ≤ 0.01; * P ≤ 0.05; NS = not significant.

*Leaf quality*

We evaluated basil quality by measuring leaf moisture content, which influences the produce's response to post-harvest processing and storage (Clarkson *et al.*, 2003), and concentration of several substances associated with the visual (chlorophylls), nutritional (antioxidant compounds), or safety (nitrate) quality.

Table 2 - Leaf concentration (on a fresh weight basis) of macro and microelements in sweet basil plants grown hydroponically for 14 days in a growth chamber under two LED light regimes with the same daily light integral (12.7 mol $m^{-2}$ $d^{-1}$): 16-hour photoperiod and 220 µmol $m^{-2}$ $s^{-1}$ PPDF (Control); 24-hour photoperiod and 147 µmol $m^{-2}$ $s^{-1}$ PPDF (CL)

| | Light regime | | ANOVA significance |
|---|---|---|---|
| | Control | CL | |
| Ca (g $kg^{-1}$) | 1.138±0.079 | 0.922±0.122 | NS |
| K (g $Kg^{-1}$) | 5.938±0.411 | 5.019±0.575 | NS |
| Mg (g $kg^{-1}$) | 0.342±0.020 A | 0.240±0.025 B | ** |
| P (g $kg^{-1}$) | 0.840±0.025 | 0.771±0.022 | NS |
| Cu (mg $kg^{-1}$) | 1.812±0.106 | 1.640±0.084 | NS |
| Mn (mg $kg^{-1}$) | 6.946±1.177 A | 3.687±0.328 B | * |
| Fe (mg $kg^{-1}$) | 16.233±1.552 | 14.061±3.366 | NS |
| Zn (mg $kg^{-1}$) | 4.185±0.268 B | 6.254±0.786 A | * |

Mean values (±SE; n = 8) flanked by different letters are significantly different at the 5% level. Significance level: *** P ≤ 0.001; ** P ≤ 0.01; * P ≤ 0.05; NS = not significant. PPFD= photosynthetic photon flux density.

In our experiments, leaf moisture content was not affected by the photoperiod (Fig. 3A). Compared to the 16-hour photoperiod, CL significantly increased the leaf concentration of total chlorophylls (+35%), flavonoids (+40%), and phenols (+44%), and the antioxidant capacity, which was measured using both FRAP (+47%) and DPPH (+63%;) assay (Fig. 3). Conversely, no significant differences in carotenoids concentration were found between the controls and CL plants (Fig. 3D) and the nitrate level was slightly but significantly reduced by CL (Fig. 3B) A significant positive correlation was found between the total antioxidant capacity measured with the two assays ($R^2$ = 0.856; n=8) and between the antioxidant capacity and the level of phenols ($R^2$=0.756 and $R^2$=0.845 for FRAP and DPPH assay, respectively) or flavonoids ($R^2$=0.956 and $R^2$=0.845).

*Electricity consumption and cost*

In our simulation, two reference light regimes were compared to CL: from 11:00 pm until 03:00 pm the following day (LR1), or from 08:00 am until 12:00 pm (LR2, Table 3).

The calculated electricity consumption for artificial lighting was 7.95 kWh $m^{-2}$ $week^{-1}$ across all three scenarios, with weekly electricity costs estimated to range from 1.30 to 1.39 € $m^{-2}$ $week^{-1}$, based on the time-of-use pricing model in Italy (Table

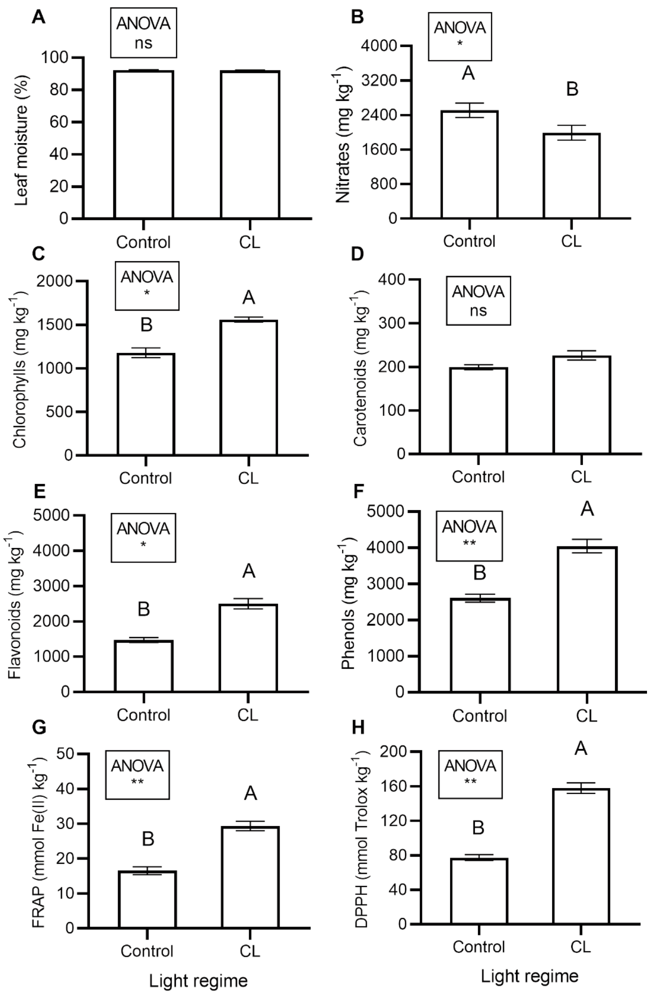


Fig. 3 - Leaf moisture content (A) and concentration (on a fresh weight basis) of nitrates (B), total chlorophylls (C), carotenoids (D), flavonoids (E), and phenols (F), and antioxidant capacity measured by FRAP (G) and DPPH (H) assay, in sweet basil plants grown hydroponically for 14 days in a growth chamber under two LED light regimes with the same daily light integral (12.67 mol m$^{-2}$ d$^{-1}$): 16-hour photoperiod and 220 µmol m$^{-2}$ s$^{-1}$ PPDF (Control); ii) 24-hour photoperiod and 147 µmol m$^{-2}$ s$^{-1}$ PPDF (CL). Mean values (±SE) of eight replicates. Significance level: *** P≤0.001; ** P≤0.01; * P≤0.05; NS = not significant. Abbreviations: PFFD, photosynthetic photon flux density; FRAP the ferric reducing ability of plasma (FRAP) assay w; DPPH, the 2,2-diphenyl-1-picrylhydrazyl assay.

3). When evaluating CL against a 16-hour light regime, we found a minor increase (+2.6%) or decrease (-4.4%) in electricity costs, which depended on the daily schedule of the latter system.

## 4. Discussion and Conclusions

According to Beaman *et al.* (2009), basil needs a dark period for optimal growth since CL may cause leaf chlorosis and necrosis, which were not observed in our experiments. In an experiment conducted in a growth chamber with the same PPFD level (250 µmol m$^{-2}$ s$^{-1}$) from LED lamps, fresh and dry biomass did not significantly differ in basil plants grown with a photoperiod of 16, 20, or 24 h d$^{-1}$, notwithstanding the large differences in DLI (14.4, 18.0, and 21.6 mol m$^{-2}$ d$^{-1}$, respectively) (Pennisi *et al.*, 2020). In contrast, in greenhouse-grown basil the supplementation of natural radiation with artificial light (from high-pressure sodium lamps) for 24 h d$^{-1}$ increased shoot DW compared to illumination for 16 or 20 h d$^{-1}$ (Islam *et al.*, 2010). Islam *et al.* (2010) used the same PPFD (150 µmol m$^{-2}$ s$^{-1}$ from HPS lamps) in the three light treatments and therefore the artificial DLIs were different (8.64, 10.80, and 12.96 mol m$^{-2}$ d$^{-1}$), and this probably accounts for the differences between their and our findings. Continuous lighting also increased the biomass of green basil microgreens compared to a 16-hour photoperiod with the same DLI (14 or 24 mol m$^{-2}$ d$^{-1}$) (Lanoue *et al.*, 2022). Positive effects of CL on plant growth were also observed in Chinese cabbage (Kang *et al.*, 2024), rocket (Proietti *et al.*, 2021), Japanese mugwort (Hata and Kawamura, 2023), and microgreens of different species (Lanoue *et al.*, 2022; Shibaeva *et al.*, 2023 b).

The significant reduction of $g_s$ detected at midnight in both treatments (Fig. 2) suggested that a circadian rhythm of stomatal movements was present in plants grown under CL (Dodd *et al.*, 2004). Similar results were found in Chinese cabbage (Kang *et al.*, 2024). In

Table 3 - Leaf concentration (on a fresh weight basis) of macro and microelements in sweet basil plants grown hydroponically for 14 days in a growth chamber under two LED light regimes with the same daily light integral (12.7 mol m$^{-2}$ d$^{-1}$): 16-hour photoperiod and 220 µmol m$^{-2}$ s$^{-1}$ PPDF (Control); 24-hour photoperiod and 147 µmol m$^{-2}$ s$^{-1}$ PPDF (CL)

| Light regime | PPFD (µmol m$^{-2}$ s$^{-1}$) | Photoperiod (h d$^{-1}$) | Daily light integral (mol m$^{-2}$ d$^{-1}$) | Lighting schedule | Costs (€ m$^{-2}$ week$^{-1}$) |
|---|---|---|---|---|---|
| LR1 | 220 | 16 | 12.7 | 11:00 pm - 03:00 pm | 1.30 |
| LR2 | 220 | 16 | 12.7 | 08:00 am - 12:00 pm | 1.39 |
| CL | 147 | 24 | 12.7 | Continuous light | 1.33 |

PPFD= photosynthetic photon fluence rate.

contrast, in soybean (Kassai, 2008) and potato (Wheeler *et al.*, 2019) grown in a growth chamber, midday $g_s$ was lower in plants grown with CL than in those grown with a photoperiod of 10-12 h $d^{-1}$.

Compared with the control, CL increased the leaf antioxidant capacity and concentration of total flavonoids and phenols and reduced the nitrate level (Fig. 3). In general, our results agree with previous findings (Table 1S). For instance, an increase in pigment concentration was observed in rocket plants grown with CL (Proietti *et al.*, 2021). In lettuce, CL improved leaf quality by reducing nitrate concentration and increasing the concentration of phenolic compounds, even when it was applied only for 1-3 days before harvest (Bian *et al.*, 2018; Yang *et al.*, 2022; Zhang *et al.*, 2021). In contrast, in several species in the Amaranthaceae, the leaf concentration of pigments and total polyphenols, and the antioxidant capacity were invariably lower in CL plants than in those grown with shorter photoperiods (Ali *et al.*, 2009).

Notwithstanding that nitrate may have positive effects on human health, nitrate intake with diet is linked with some health risks (Karwowska and Kononiuk, 2020). Therefore, the nitrate level in drinking water and food is strictly regulated and some agronomic practices have been proposed to reduce nitrate content in vegetables, which represent the primary source of nitrate in the human diet (Colla *et al.*, 2018). Sweet basil is prone to accumulate high levels of nitrate (Corrado *et al.*, 2020). The CL-induced reduction in leaf nitrate concentration observed in our study agrees with previous findings in other crops (Bian *et al.*, 2016; Proietti *et al.*, 2021; Shibaeva *et al.*, 2023 b). It is known that light regulates nitrate reductase (Lillo and Appenroth, 2001); CL can increase the activity of nitrate reductase (NR) and consequently the reduction of nitrate in plant leaves. In lettuce, the reduction of leaf nitrate concentration induced by short-term (two days) CL was associated with increased activity and gene expression of NR and nitrite reductase (Bian *et al.*, 2016, 2018). In contrast, CL augmented leaf nitrate levels in purslane (He *et al.*, 2023) and this was associated with a reduction in NR, compared with a 12-hour photoperiod.

Continuous illumination can increase the generation of reactive oxygen species (ROS) and induce oxidative stress (Haque *et al.*, 2015; Huang *et al.*, 2019; Liu and Liu, 2024). The balance between ROS production and the plant's ability to scavenge these molecules largely determines the extent of CL-induced plant damage (Kumar *et al.*, 2022). If this balance is achieved, injury-free production is possible under CL, with the additional improvement of produce quality resulting from a higher content of antioxidants (e.g. phenolics) and other health-promoting compounds, as was found in this and other works (Bian *et al.*, 2016; Hata and Kawamura, 2023). As such, applying CL for indoor cultivation of short-cycle crops can be considered an eustressor capable of promoting crop yield and improving produce quality (Vázquez-Hernández *et al.*, 2019).

For the same DLI, the use of CL can markedly decrease the number (-33% compared to the 16-hour photoperiod, in this work) and the installation costs of light fixtures. Moreover, in countries that implement a time-of-use pricing model of the electricity market, the operational costs of lighting are lower during off-peak periods. Electricity prices are generally lower early in the day, overnight, and during the weekends and holidays.

For basil production in vertical farms, illumination is generally operated for 16 continuous hours per day, switching on LEDs during the nighttime, when energy prices are lower than in the daytime (Avgoustaki and Xydis, 2021). According to our simulation, CL would slightly increase or decrease electricity costs compared to 16-hour illumination, depending on the daily schedule of the standard lighting regime (Table 3).

For microgreens production in vertical farms, compared to a 16-hour photoperiod, CL reduced electricity costs (expressed per unit of fresh biomass) by 8% to 38%, when DLIs were the same (Lanoue *et al.*, 2022). However, Lanoue and colleagues (Lanoue *et al.*, 2022) did not clarify when the light was switched on in the control treatment.

Vertical farms use powerful air conditioning systems to regulate air temperature and humidity for optimal crop production (Zhu and Marcelis, 2023). Continuous illumination at relatively low PPFD reduces the heat generated by LED lamps and the moisture released through plant transpiration, thus reducing the demand for air cooling and dehumidification (Cai *et al.*, 2024). In addition, CL could provide beneficial effects for pest and disease management. For instance, CL was found to suppress the sporulation of *Peronospora belbahrii,* which is the causal organism of basil downy mildew, one of the most destructive diseases of sweet basil (Radetsky *et al.*, 2020). Continuous light also reduced whiteflies' infestation (*Trialeurodes vaporariorum)* in greenhouse roses (Johansen, 2009).

This work confirms the excellent adaptability of sweet basil to vertical farms since abundant yield can be achieved in a couple of weeks.

Continuous illumination at relatively low light intensity can be used to reduce the capital costs for the lighting system with no critical effects on crop yield and electricity costs in vertical farms. Leaf quality was improved by continuous lighting due to a lower nitrate level and higher antioxidant capacity and concentration of total flavonoids and phenols.

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